ABSTRACT OF THE DISCLOSURE

A method is disclosed for modifying an oligonucleotide, which method has application to the detection of a polynucleotide analyte. An oligonucleotide is reversibly hybridized with a polynucleotide, for example, a polynucleotide analyte, in the presence of a 5′-nuclease under isothermal conditions. The polynucleotide analyte serves as a recognition element to enable a 5′-nuclease to cleave the oligonucleotide to provide (i) a first fragment that is substantially non-hybridizable to the polynucleotide analyte and (ii) a second fragment that lies 3′ of the first fragment (in the intact oligonucleotide) and is substantially hybridizable to the polynucleotide analyte. At least a 100-fold molar excess of the first fragment and/or the second fragment are obtained relative to the molar amount of the polynucleotide analyte. The presence of the first fragment and/or the second fragment is detected, the presence thereof indicating the presence of the polynucleotide analyte. The method has particular application to the detection of a polynucleotide analyte such as DNA. Kits for conducting methods in accordance with the present invention are also disclosed.